The Influence of Schizophyllum commune on Asthma Severity

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The influence of *Schizophyllum commune* on the severity and exacerbation of asthma

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Conflict of interest

The all authors declare that they have no competing interests that might be perceived to influence the results and discussion reported in the present manuscript.

A short running head

Impact of *S. commune* on asthma

Abstract

The sensitization and exposure to fungal allergens have been reported to be associated with asthma. The aim of this study was to clarify the impact of sensitization to *Schizophyllum commune* (*S. commune*) on the severity and exacerbations of asthma.

Ninety-two patients with asthma of various levels of severity (mild (n=18), moderate (28), and severe (46)) and exacerbation severity (moderate (n=43) and severe (6)), were retrospectively examined with regard to fungal sensitization such as specific IgE or intradermal skin reactions against *S. commune* and other common allergens. We also classified the patients into 3 groups; 1) 3 or more asthma attacks during the past 1 year (F-BA) (n=29), 2) one or two asthma attacks (NF-BA) (n=20), and 3) no asthma attack (C-BA) (n=43).

The positive rate of late cutaneous reactions to *S. commune* was higher in patients with severe asthma (41.2%) than moderate (26.1%) and mild asthma (6.7%), and was significantly different among the three groups ($p<0.05$). Though the ratio did not show significant difference between the patients having a severe (83.3%) and moderate (36.1%) exacerbation, it was higher in F-BA (60.9 %) than in NF-BA (21.1%) and C-BA patients (10.0%), and it was significantly different among the three groups ($p=0.0002$). A multivariate analysis identified positive results for late phase skin

reactions to *S. commune* and the age of the patients as an independent determinant of asthma severity, and the skin results and %FVC an independent determinant of exacerbation frequency.

This study demonstrated that *S. commune* is an environmental fungus that appears to enhance both the severity of asthma and the exacerbation frequency.

Key words

Asthma severity, severity of exacerbation, exacerbation frequency, basidiomycetous fungus, fungal sensitization, *Schizophyllum commune*

Introduction

Throughout the Global Initiative for Asthma (GINA) Science Committee report in 2009 [1], emphasis is placed on the concept that the goal of asthma treatment is to achieve and maintain clinical control. Each asthmatic patient should be assessed to establish their level of asthma control according to the 5 items of clinical manifestations, and patients are then assigned to one of five "treatment steps" in accordance with the management approach based on the level of asthma control. Furthermore, as assessment of future risk, frequent exacerbations in past year are listed as the features that were associated with an increased risk of adverse events in the future. Because patients with well-controlled asthma are less likely to experience exacerbations than those whose asthma is not well-controlled, preventing any exacerbation of asthma is an important clinical issue in asthma control.

It is well-known that fungal exposure is associated with exacerbations. Recently the sensitization and exposure to fungal allergens such as *Aspergillus, Alternaria, Penicillium, Cladosporium* and *Trichophyton* have been reported to be associated with asthma exacerbations and severity [2], and the frequency of exacerbations, treatment requirements, and admission to intensive care for asthma has been found to be associated with skin-test reactivity to one or more fungi [3]. Thus the investigations to

identify environmental fungi are considered to be part of initial assessment to enable the use of avoidance strategies in asthma management [4].

From the results of our series of studies [5, 6] on patients with allergic fungal respiratory diseases, we have focused on the possible role of basidiomycetous (BM) fungi especially *Bjerkandera adusta* [7] (*B. adusta*; the major cause of "allergic fungal cough" in Japan) as a fungal aeroallergen [8]. We recently encountered two cases of asthma caused by *Schizophyllum commune* (*S. commune*, "suehirotake" in Japanese) [9]. Although this fungus, one of the BM fungi, is well-known as a causative fungus of mucoid impaction of the bronchi (MIB) [10], allergic bronchopulmonary mycosis (ABPM) [11,12] and allergic fungal sinusitis (AFS) [13], the influence of the sensitization to the fungus in asthmatics has not yet been reported.

Therefore we compared the allergological findings of the asthmatics and reported the impact of sensitization to *S. commune* on the severity and the exacerbations of asthma.

Methods and Materials

The medical records of 92 patients with asthma referred to Saiseikai Kanazawa Hospital between August and October 2009, who had not been treated with maintenance oral

corticosteroids, were collected and reviewed retrospectively. In this study, the severity of asthma in the patients had been already classified based on their clinical features and lung function in accordance with the 2006 GINA guidelines [14]; while the previous classification was recommended only for research purposes by GINA 2009. The asthma exacerbation severity was classified according to the recent recommended definitions provided by the Task Force [15]; wherein “severe exacerbations” were classified as events that usually require hospitalization/emergency room (ER) visits and/or the use of systemic corticosteroids for at least 3 days (Acute care settings), and “moderate exacerbation” as an event that leads to an increase in the need for existing asthma therapy not including systemic corticosteroids or a change in asthma symptoms/lung function over a period of at least 2 days, not warranting hospital admission (Community settings).

Because the classification of the frequency of exacerbation has not yet been clearly defined, we classified the patients who had experienced exacerbations which required intravenous corticosteroid therapies in spite of adequate interval control into 3 groups according to the frequency of the episodes as an assessment of exacerbation tendency; 1) patients who had experienced exacerbations 3 times or more during the past 1 year (frequent group; F-BA), 2) patients who had experienced one or two exacerbations

(non-frequent group; NF-BA), and 3) the other patients who had never experienced such exacerbations (controlled group; C-BA).

The following information was collected from the medical records of all of the patients: name, date of birth, gender, smoking habits, and the results of examinations such as blood tests, chest radiographs, and pulmonary function tests. And for the measurements of allergic status, the allergological tests such as intradermal skin test and serological test had been performed at least 3 months after the latest exacerbation. Patients with asthma of each severity, each exacerbation severity, and each exacerbation frequency were compared in a cross-sectional manner with regard to gender, age, FEV1, and allergological characteristics.

This retrospective study was approved by the Institutional Review Board (reference number 2009004), and written informed consent was obtained from each of the 92 patients.

Preparation of the antigenic solution

Aspergillus, Alternaria, Candida: These antigens were commercially available (Torii Pharmacy, Tokyo, Japan).

B. adusta, S. commune: One liter of Sabouraud dextrose broth in 3 liter flasks was

sterilized. Five mL of a *B. adusta* (NBRC 4983) or *S. commune* spore suspension (10^5 spores per mL) in sterile physiological saline from 14-day-old Sabouraud dextrose agar culture were used to inoculate the flask. The flask was shaken at 150 rpm in a 25°C rotary shaker incubator for 14 days. Mycelia were separated by filtration and centrifuged. The supernatants were dialyzed against 5 mM ammonium bicarbonate and lyophilized.

Allergological tests

Intradermal skin test

An antigenic solution (polysaccharide) was injected intradermally with a tuberculin syringe (0.02 mL, 1 mg/mL) to assess the skin response to the solution. The results were judged to be positive when the longer axis of the flare exceeded 9 mm at 15 minutes (Immediate phase), and 10 mm at 8 hrs (late phase) after the injection.

The results of the late skin test were checked and recorded by each patient at 8 hours after the tests. The next day, we telephoned the patients and completed the examination.

Serological test

House-dust, Mite, Aspergillus, Alternaria, Penicillium, Cladosporium, Candida

Trichophyton: Allergen-specific IgE antibodies were detected with a capsulated hydrophilic carrier polymer radioallergosorbent test fluoroenzyme immunoassay (Phadia, Uppsala, Sweden) at an external laboratory (SRL, Tokyo, Japan)

Schizophyllum commune: The Phadia (previously Pharmacia) CAP system was used to quantify specific IgE levels (Phadia Ltd, Uppsala, Sweden) [16]. A positive test was taken as a measurement > 0.35 units of allergen (UA)/mL.

Statistical analysis

Variables are expressed as the mean (SD) unless otherwise stated. For comparison of multiple groups, analysis of variance (ANOVA) followed by Fisher protected least significant difference post hoc test was used for parametric data, when significant difference was found. For nonparametric data, the Kruskal-Wallis test followed by the Mann-Whitney *U* test was applied instead. The χ^2 test was used for categorical data. Logistic regression analysis was performed to test for independent effects of age, smoking habit, lung functions, and positive late phase skin reactions against *S. commune* on asthma severity and exacerbation severity. A multiple regression analysis was performed on exacerbation frequency. Analyses were performed by using a statistical software package (StatView; SAS Institute Inc;

Cary, NC). A p value of < 0.05 was considered statistically significant.

Results

The clinical records of the 92 asthmatics were gathered and analyzed. Chest and sinus radiographs were normal in all patients. The results of the late phase positive skin reaction tests were obtained from 57, 72, and 57 patients for *Aspergillus, S. commune,* and *B. adusta,* respectively.

Analysis for asthma severity (Table.1)

Table 1 shows the characteristics and results of the three groups of asthma severity. Patients with mild and moderate severities of asthma were significantly younger than the patients with severe asthma. The smoking habits did not differ among the groups. The FVC (% predicted), FEV1 (% predicted) and the FEV1/FVC ratio (%) in mild and moderate asthmatics were significantly higher than those in severe asthmatics ($p < 0.05$, $p < 0.01$, and $p < 0.01$, respectively). The total IgE levels and the positive rate for a specific IgE did not differ among these groups.

The positive skin reaction rates did not differ among these groups except for the late phase skin reaction against *S. commune* ($p < 0.05$). The results of an investigation of the serum *S. commune*-specific IgE levels were available for 12 patients, and the value was

not elevated in any of them.

A multiple logistic regression analysis was performed by including age and the late phase skin reaction against *S. commune* as possible predictors of asthma severity. The probability of severe vs mild and moderate asthma was found to be significantly associated with age and positive late phase skin reaction against *S. commune* (Table.4).

Analysis for exacerbation severity (Table.2)

In this analysis, 43 patients who had never experienced any exacerbations were excluded. Table 2 shows the characteristics and results of two groups divided by their exacerbation severity. Gender and age were well matched within the each group. Current smoker showed predominance in the severe exacerbation group. The FVC (% predicted) and FEV1 (% predicted) in the moderate exacerbation group were significantly higher than those in the severe exacerbation group.

The number of patients from whom we had obtained the results of the late phase positive skin reaction tests for each fungus was shown in Table 2. The positive of the late phase skin reaction against *S. commune* (83.3%) was significantly higher in the severe exacerbation group than in the moderate exacerbation group ($p < 0.01$).

A multiple logistic regression analysis was performed by including smoking habits, lung functions, and the late phase skin reaction against *S. commune* as possible

predictors of exacerbation severity. The probability of the severe vs moderate exacerbation was not demonstrated in this analysis (Table 4).

Analysis for exacerbation frequency (Table.3)

Table 3 shows the characteristics and results of three groups divided based on their exacerbation frequency. The age distribution, the gender of the population, and smoking habits did not differ significantly among three groups. The FVC (% predicted) and FEV1/FVC ratio (%) in the C-BA and NF-BA groups were significantly higher than those in the F-BA group ($p < 0.05$, and $p < 0.01$, respectively). The total IgE levels and the rate of positive specific IgE did not differ significantly among the 3 groups. The rate of positive skin reactions did not differ among these groups except for the late phase skin reaction against *S. commune* ($p < 0.001$). Because the %FVC and late skin reaction against *S. commune* were adopted as possible predictors of the exacerbation frequency according to a stepwise analysis among the 3 items, thereafter multiple logistic analyses were performed by including the two variables except FEV1/FVC ratio (%). The probability of asthma exacerbation frequency was found to be significantly associated with %FVC and positive late phase skin reaction against *S. commune* ($p < 0.0001$) on the basis of a multiple regression analysis (Table 4).

Discussion

Bronchial asthma is a chronic inflammatory disease of the airways, which may worsen due to numerous extrinsic factors such as continuous exposure to fungal agents. The factors that influence the risk of asthma have been divided into those that cause the development of asthma and those that trigger asthma symptoms. It is known that fungi involved in outdoor allergens are important as environmental factors, and our recent studies suggested that *S. commune* is an important candidate organism acting as a causative fungal antigen of bronchial asthma [9] similar to *Trichophyton* species [17-20]. Therefore, we retrospectively examined whether an environmental fungus such as *S. commune* could enhance the clinical expression of asthma.

Asthma severity has recently been classified on the basis of the intensity of treatment required to achieve good asthma control in subjects with “mild asthma” and “severe asthma”. The patient’s asthma severity will be established based on a combination of their current level of symptoms and their current maintenance treatment step in future studies; however, in this retrospective study, the severity of all asthmatics had been classified based on their daily medication regimen and response to treatment, as described in the GINA 2006 guidelines for research purposes [1]. Matsuoka et al.

reported a strong association of the allergen with the severity of asthma and the establishment of sensitization using serum radioallergosorbent assays for IgE antibodies to Trichophyton [19], in contrast to their report, our present results demonstrated no association of serum specific-IgE antibodies to Trichophyton with the severity of asthma. The presence of Trichophyton infection has been suggested to be an important determinant of sensitization to Trichophyton; however in our study, there were no significant differences in the frequencies of fungal skin infections among the asthma severities. Therefore, it is impossible that fungal skin infections may affect the detection frequency of specific IgE against Trichophyton in our study. The present study revealed that there were no significant differences in the positive results for the immediate and late cutaneous reactions to 3 well-known environmental fungi (Aspergillus, Alternaria, and Candida) among the asthma severities [21]. The reason for this result is unclear, but it might be related to geographic differences in the frequency of sensitization.

Asthma exacerbations are considered to be caused by a variety of factors, such as allergens, viral infections, pollutants, drugs, and also fungi. Therefore, reducing a patient's exposure to risk factors will improve the control of their asthma and reduce their need for medication [22]. Each patient was assigned to one of five treatment steps depending on their current level of control, and treatment was adjusted in a continuous

cycle which involved: assessing asthma control, treating to achieve control, monitoring to maintain control, and by changes in their asthma control status. Because asthma is a variable disease, treatment had to be adjusted periodically in response to loss of control, as indicated by either worsening symptoms or the development of exacerbation. Most recently, an American Thoracic Society (ATS)/European Respiratory Society (ERS) consensus report aimed to standardize the definitions of exacerbations for clinical trials, and defined as a "severe exacerbation" and "moderate exacerbation" [15]. Our current study revealed a significant difference in the smoking habit, lung function, and the rate of positive late phase skin reactions against *S. commune,* but the probability of the severe vs moderate exacerbation was not demonstrated based on a multiple regression analysis, thus suggesting that many other factors may be associated with the variability in the severity of exacerbation.

In the GINA 2009, a history of frequent exacerbations in the past year was emphasized from the aspect of assessment of future risk, and was listed as one of the features that were associated with an increased risk of adverse events in the future. The severity of the each event of the asthma attack, which reflects exacerbation rather than just transient loss of asthma control, may be affected by the magnitude of the response against viral infection or by the some other factors, such as physical condition or severe

weather. We therefore assumed that the so-called “exacerbation tendency” of asthma was represented by the frequency of exacerbations rather than by the severity of each exacerbation.

A multivariate analysis identified positive results for late phase skin reactions to *S. commune* and age of the patients as an independent determinant of asthma severity, and the skin results and %FVC as that of exacerbation frequency, but such a trend was not found for other allergens such as house-dust, Aspergillus, Alternaria, Penicillium, and Cladosporium [23].

There is emerging evidence that the innate immune response in asthma is impaired, airway inflammation may modulate the response to triggers and genetic polymorphisms may confer an increased risk of exacerbations [24]. Because it has been already suggested that the presence of eosinophilic inflammation in the bronchial wall promotes the development of an exacerbation in combination with a viral infection, a clinical research study concerning the eosinophil response in the bronchial wall to *S. commune* or to the secretory products of this fungus will also be important to elucidate the mechanisms underlying the exacerbation of asthma in the future.

From the results of the present study, *S. commune*, a BM fungus, is considered to be one of the environmental fungi which have a potential role in enhancing the severity

of asthma and asthma exacerbation via sensitization to the fungus. *S. commune* colonizes rotting wood, and it is distributed throughout the world. Therefore, not only pharmacologic intervention [25, 26] but also careful environmental management [12] may positively help to prevent the development of asthma, asthma symptoms, and asthma exacerbations by either reducing or eradicating such antigen exposure.

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Tables

Table 1 Characteristics and results of three groups of asthma severity

Table 2 Characteristics and results of two groups of asthma exacerbation severity

Table 3 Characteristics and results of three groups of asthma exacerbation frequency

Table 4 Results of multiple logistic regression analysis

The analysis was performed by including age, *Trichophyton*-specific IgE, and the late phase skin reaction against *S. commune* as possible predictors of the severity and exacerbation of asthma.

Table 1 The characteristics and results of three groups of asthma severity

		Asthma severity			
Characteristics		Mild	Moderate	Severe	p Value
Patients, No		18	28	46	
Gender, M/F (F %)		6 / 12 (66.7)	17 / 11 (39.3)	27 / 19 (41.3)	NS
Age, yr		54.7 ± 15.2	56.5 ± 15.8	64.9 ± 12.1	**p < 0.01**
Smoking habit (never, ex, current)		16 / 1 / 1	24 / 1 / 3	33 / 6 / 7	NS
FVC, % predicted		114.9 ± 14.2	113.6 ± 18.2	100.8 ± 25.7	**p < 0.05**
FEV1, % predicted		108.6 ± 5.4	105.8 ± 4.3	91.4 ± 3.4	**p < 0.01**
FEV1 / FVC ratio (%)		79.0 ± 9.1	76.0 ± 9.9	65.3 ± 17.5	**p < 0.01**
IgE, IU / ml		262.0 ± 222.7	301.8 ± 178.6	611.2 ± 140.9	NS
Positive specific IgE					
House-dust		9 (50)	11 (39.3)	14 (30.4)	NS
Mite		8 (44.4)	7 (25.0)	14 (30.4)	NS
Aspergillus		1 (5.6)	2 (7.1)	4 (8.7)	NS
Alternaria		0 (0)	1 (3.6)	2 (4.4)	NS
Penicillium		0 (0)	2 (7.1)	4 (8.7)	NS
Candida		2 (11.1)	2 (7.1)	13 (28.3)	NS
Trichophyton		1 (5.6)	3 (10.7)	5 (10.9)	NS
S. commune		0 / 1 (0)	0 / 4 (0)	0 / 7 (0)	NS
Positive skin reaction					
Aspergillus	Immediate	1 (5.6)	4 (14.3)	6 (13.0)	NS
	Late	1 / 11 (9.1)	2 / 21 (9.5)	3 / 25 (12.0)	NS
S. commune	Immediate	1 (5.6)	6 (21.4)	13 (28.3)	NS
	Late	1 / 15 (6.7)	6 / 23 (26.1)	14 / 34 (41.2)	**p < 0.05**
B. adusta	Immediate	0 (0)	2 (7.1)	6 (13.4)	NS
	Late	5 / 13 (38.5)	5 / 19 (26.3)	9 / 25 (36.0)	NS
Alternalia	Immediate	1(5.9)	2 (7.1)	1 (2.2)	NS
Candida	Immediate	10 (58.8)	16 (57.1)	30 (65.2)	NS
Skin fungal infection		7 (38.9)	14 (50.0)	15 (32.6)	NS
HbA1c		5.3 ± 0.8	5.2 ± 1.2	5.5 ± 0.7	NS

Table 2 The characteristics and results of two groups of asthma exacerbation severity

		Exacerbation severity		
Characteristics		Moderate	Severe	p Value
Patients, No		43	6	
Gender, M/F (F %)		20 / 23 (53.5)	5 / 1 (16.7)	NS
Age, yr		62.8 ± 13.6	67 ± 10.2	NS
Smoking habit (never, ex, current)		34 / 5 / 4	2 / 1 / 3	**p < 0.01**
FVC, % predicted		108.3 ± 17.4	81.7 ± 29.8	**p < 0.05**
FEV1, % predicted		98.4 ± 23.6	63.5 ± 34.3	**p < 0.05**
FEV1 / FVC ratio (%)		70.2 ± 14.0	55.6 ± 18.1	NS
IgE, IU / ml		365.9 ± 596.5	1792.2 ± 3095.3	NS
Positive specific IgE				
House-dust		13 (30.2)	2 (33.3)	NS
Mite		13 (30.2)	2 (33.3)	NS
Aspergillus		2 (4.7)	1 (16.7)	NS
Alternaria		1 (2.3)	1 (16.7)	NS
Penicillium		2 (4.7)	1 (16.7)	NS
Candida		10 (23.3)	3 (50.0)	NS
Trichophyton		2 (4.7)	1 (16.7)	NS
S. commune		0 / 6 (0)	0 / 2 (0)	NS
Positive skin reaction				
Aspergillus	Immediate	2 (4.7)	1 (16.7)	NS
	Late	2 / 28 (7.1)	1 / 4 (25.0)	NS
S. commune	Immediate	8 (18.6)	3 (50.0)	NS
	Late	13 / 36 (36.1)	5 / 6 (83.3)	**p < 0.05**
B. adusta	Immediate	5 (11.6)	0 (0)	NS
	Late	10 / 28 (35.7)	2 / 4 (50.0)	NS
Alternalia	Immediate	1 (2.3)	0 (0)	NS
Candida	Immediate	29 (67.4)	4 (66.7)	NS
Skin fungal infection		15 (34.9)	3 (50.0)	NS
HbA1c		5.6 ± 0.9	5.3 ± 0.3	NS

Table 3 The characteristics and results of three groups of asthma exacerbation frequency

		Exacerbation frequency			
Characteristics		Controlled	Non-frequent	Frequent	p Value
Patients, No		43	20	29	
Gender, M/F (F %)		25 / 18 (41.9)	9 / 11 (55.0)	16 / 13 (44.8)	NS
Age, yr		56.9 ± 15.3	62.0 ± 14.6	64.3 ± 12.3	NS
Smoking habit (never, ex, current)		37 / 2 / 4	17 / 1 / 2	19 / 5 / 5	NS
FVC, % predicted		112.9 ± 17.8	110.5 ± 16.0	97.2 ± 29.0	**p < 0.05**
FEV1, % predicted		104.1 ± 18.2	100.7 ± 19.0	90.8 ± 32.0	NS
FEV1 / FVC ratio (%)		75.5 ± 10.7	72.2 ± 13.3	64.3 ± 19.6	**p<0.01**
IgE, IU / ml		331.0 ± 396.4	318.4 ± 724.0	703.1 ± 1490.2	NS
Positive specific IgE					
House-dust		19 (44.2)	5 (25.0)	10 (34.5)	NS
Mite		14 (32.6)	5 (25.0)	10 (34.5)	NS
Aspergillus		4 (9.3)	1 (5.0)	2 (6.9)	NS
Alternaria		1 (2.3)	1 (5.0)	1 (3.45)	NS
Penicillium		3 (7.0)	1 (5.0)	2 (6.9)	NS
Candida		4 (9.3)	4 (20.0)	9 (31.0)	NS
Trichophyton		6 (14.0)	1 (5.0)	2 (6.9)	NS
S. commune		0 / 4 (0)	0 / 0 (0)	0 / 8 (0)	NS
Positive skin reaction					
Aspergillus	Immediate	8 (18.6)	0 (0)	3 (10.3)	NS
	Late	3 / 25 (12.0)	0 / 15 (0)	3 / 17 (17.7)	NS
S. commune	Immediate	9 (21.0)	5 (25.0)	6 (21.0)	NS
	Late	3 / 30 (10.0)	4 / 19 (21.1)	14 / 23 (60.9)	**p<0.001**
B. adusta	Immediate	3 (7.0)	3 (15.0)	2 (6.9)	NS
	Late	7 / 25 (28.0)	4 / 17 (23.5)	8 / 15 (53.3)	NS
Alternalia	Immediate	3 (7.1)	1 (5.0)	0 (0)	NS
Candida	Immediate	23 (54.8)	11 (55.0)	22 (75.9)	NS
Skin fungal infection		18 (41.9)	8 (40.0)	10 (34.5)	NS
HbA1c		5.1 ± 0.9	5.5 ± 0.9	5.6 ± 0.8	NS

Table 4 Results of multiple logistic regression analysis

Asthma severity

	Severe vs. moderate and mild asthma		
Variables	OR	95%CI	p Value
Age	1.04	1.00-1.08	**p < 0.05**
Positive late skin reaction against *S. commune*	1.82	1.06-3.24	**p < 0.05**

Exacerbation severity

	Severe vs. moderate exacerbation		
Variables	OR	95%CI	p Value
Smoking habit	8.53	0.84-14.50	0.11
FEV1, % predicted	0.01	0.91-1.00	0.08
Positive late skin reaction against *S. commune*	9.63	0.93-18.17	0.11

Exacerbation frequency

R^2=0.25		Standardized Coefficients	
Variables	PRC	β	p Value
%FVC	-0.01	-0.23	**p < 0.05**
Positive late skin reaction against *S. commune*	0.41	0.43	**p < 0.0001**

CI: cofidence interval; OR: odds ratio

PRC: partial regression coefficient

The analysis was performed by including age, *Trichophyton*-specific IgE, and the late phase skin reaction against *S. commune* as possible predictors of the severity and exacerbation of asthma.